Functional validation of CoQ_{10} deficiency fibroblast model with COQ7 mutations

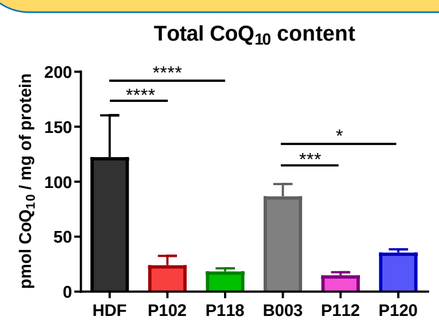
Manuel Torralba-Carnerero, Ana Belén Cortés-Rodríguez, María Alcázar-Fabra, Carmine Staiano, Ana Sánchez-Cuesta, Manuel Ballesteros-Simarro, María Victoria Cascajo-Almenara, Carlos Santos-Ocaña

Centro Andaluz de Biología del Desarrollo, Universidad Pablo de Olavide, 41013 Sevilla, Spain.

Coenzyme Q_{10} (CoQ_{10}) deficiency syndrome comprises a heterogeneous group of **mitochondrial disorders** characterized by a decrease in CoQ_{10} content in cells and tissues. Primary CoQ_{10} deficiencies are rare genetic conditions caused by mutations in COQ genes, whose encoded proteins are directly linked to the final biochemical pathway of CoQ biosynthesis. **COQ proteins** are disposed forming a mitochondrial complex in which **COQ7** is included, catalysing the hydroxylation of DMQ_{10} into $DMeQ_{10}$, one of the latest steps of the CoQ biosynthetic pathway. Here we present **four clinical cases** of primary CoQ_{10} deficiency, which is presumably caused by ***Coq7* mutations**. The motivation for this work is to **validate** it in a cellular model based on primary cultures from patients' skin fibroblasts, in order to complete the previously started **molecular diagnosis** by whole-exome sequencing.

RESULTS

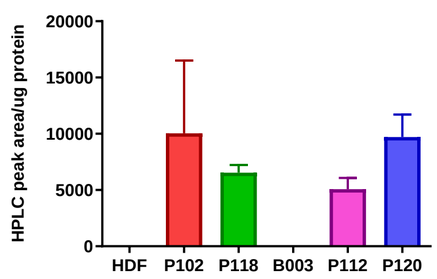
CoQ_{10} Deficiency

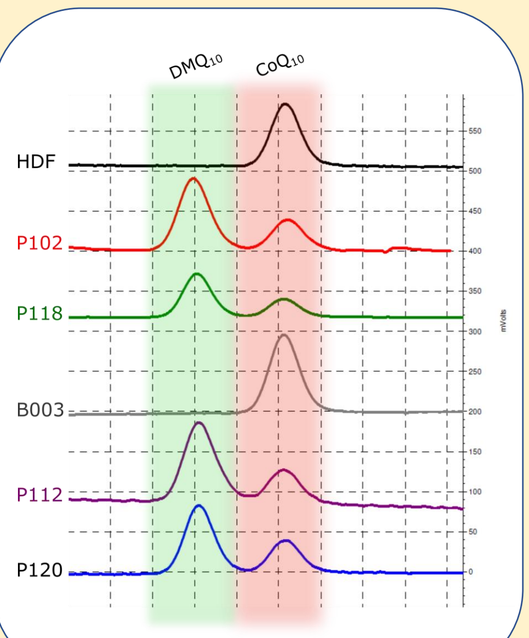


One-way ANOVA, Sidak's multiple comparisons test

p value: <0.0001(****); =0,001(***); <0,05(*)

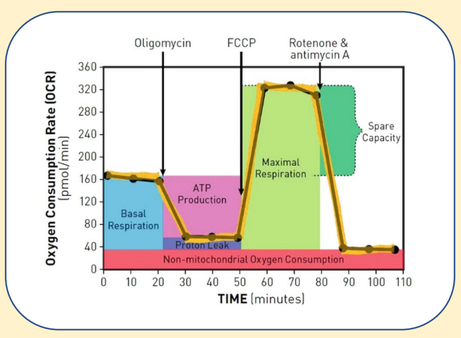
DMQ_{10} Accumulation



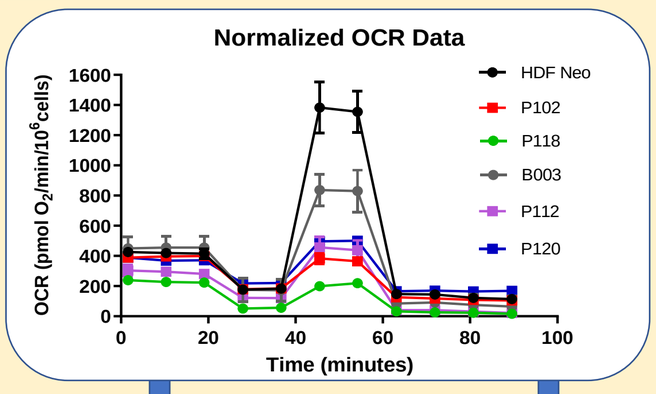


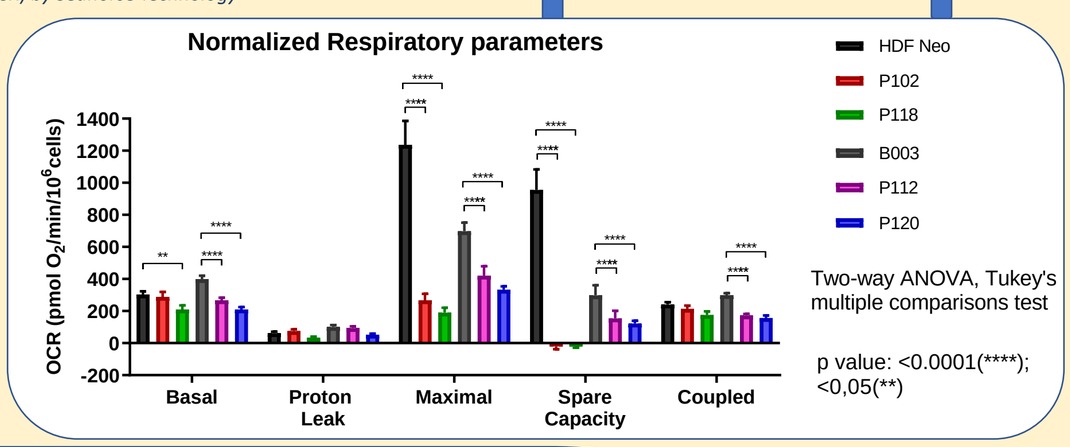
Control	Patients' cells	
HDF	P102	P118
B003	P112	P120

Mitochondrial Respiration is affected



Representative Oxygen Consumption Rate (OCR) by Seahorse Technology



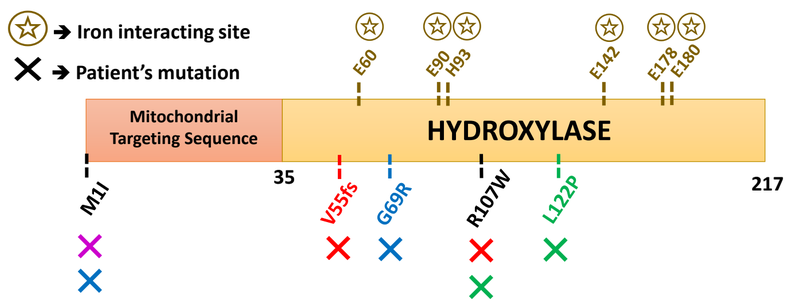


Two-way ANOVA, Tukey's multiple comparisons test

p value: <0.0001(****); <0,05(**)

COQ7 Protein

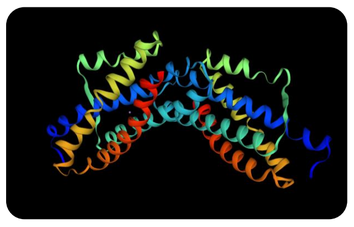
COQ7 Domains



Patients' Mutations

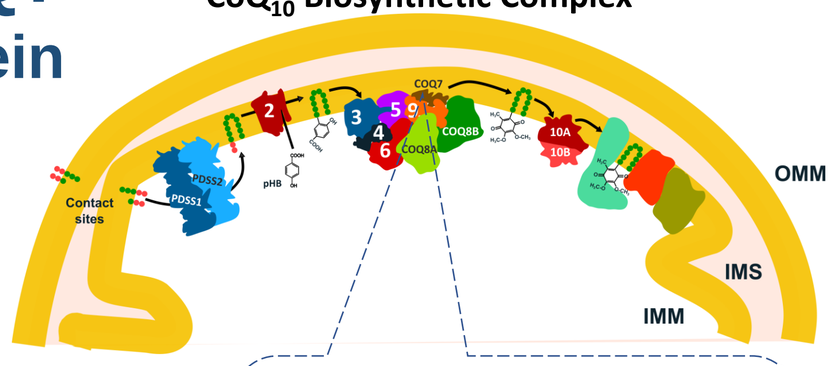
	Maternal	Paternal
P102	Val55fs	Arg107Trp
P112	Met1Ile	Met1Ile
P118	Arg107Trp	Leu122Pro
P120	Met1Ile	Gly69Arg

3D Model

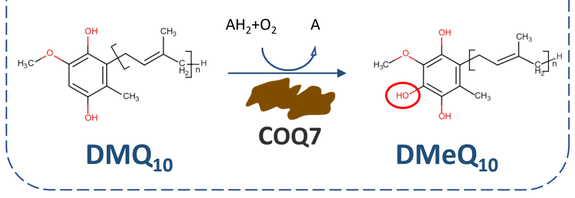


COQ7 protein

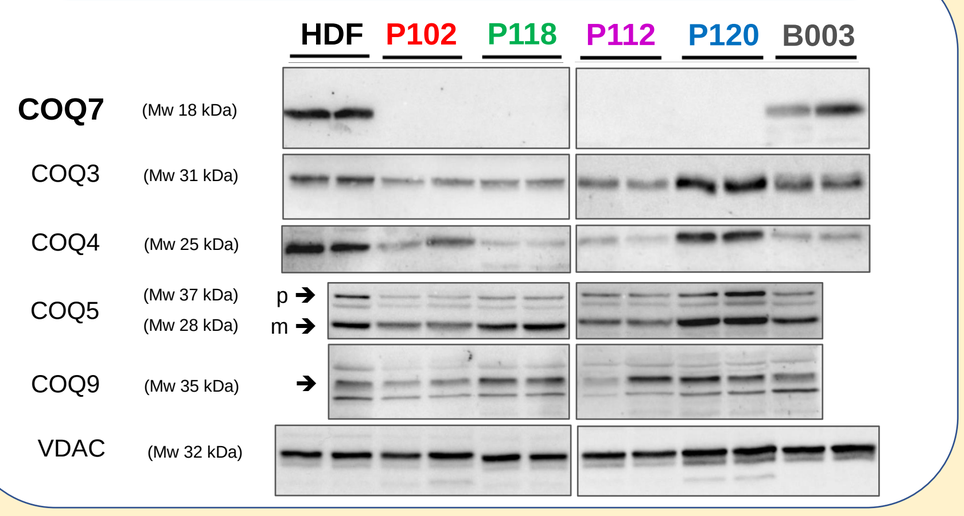
CoQ_{10} Biosynthetic Complex



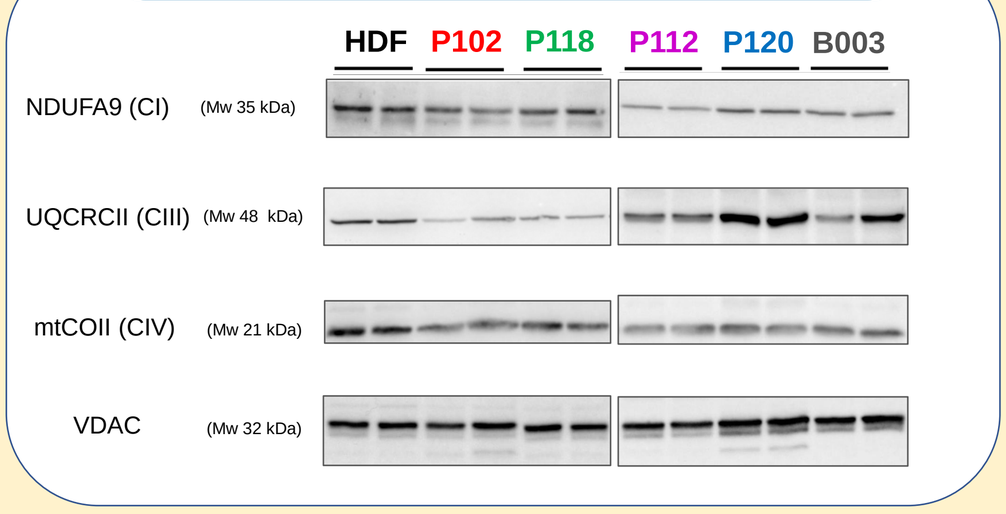
Hydroxylase Function



COQ Proteins



Respiratory Chain Proteins



CONCLUSIONS

- ✓ Confirmed **CoQ_{10} deficiency** in patients' fibroblast
- ✓ **Reaction catalysed by COQ7** is **impaired** since patients **accumulate DMQ_{10}**
- ✓ ***Coq7* mutation** affects to other **COQ and Respiratory Chain proteins expression**, **CoQ_{10}** levels and **mitochondrial respiration**
- ✓ Our data **support the previous diagnosis** obtained by exome analysis, proving that in these clinical cases, **the CoQ_{10} deficiency is being produced by the absence of COQ7 protein**

REFERENCES

1. Alcázar-Fabra, M., Navas, P., and Brea-Calvo, G. (2016) Coenzyme Q biosynthesis and its role in the respiratory chain structure. Biochim. Biophys. Acta - Bioenerg. 1857, 1073–1078
2. Delia Yubero, Raquel Montero, Carlos Santos-Ocaña, Leonardo Salviati, Placido Navas & Rafael Artuch (2018) Molecular diagnosis of coenzyme Q10 deficiency: an update, Expert Review of Molecular Diagnostics, 18:6, 491-498,
3. Stenmark P, Grünler J, Mattsson J, Sindelar PJ, Nordlund P, Berthold DA. A new member of the family of di-iron carboxylate proteins. Coq7 (clk-1), a membrane-bound hydroxylase involved in ubiquinone biosynthesis. J Biol Chem. 2001 Sep 7;276(36):33297-300.

Manuel Torralba Carnerero